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Genome-wide scan of job-related exhaustion with three replication studies implicate a susceptibility variant at the *UST* gene locus

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Job-related exhaustion is the core dimension of burnout, a work-related stress syndrome that has several negative health consequences. In this study, we explored the molecular genetic background of job-related exhaustion. A genome-wide analysis of job-related exhaustion was performed in the GENMETS subcohort ($n = 1256$) of the Finnish population-based Health 2000 study. Replication analyses included an analysis of the strongest associations in the rest of the Health 2000 sample ($n = 1660$ workers) and in three independent populations (the FINRISK population cohort, $n = 10\,753$; two occupational cohorts, total $n = 1451$). Job-related exhaustion was ascertained using a standard self-administered questionnaire (the Maslach Burnout Inventory (MBI)-GS exhaustion scale in the Health 2000 sample and the occupational cohorts) or a single question (FINRISK). A variant located in an intron of *UST,* uronyl-2-sulfotransferase (rs13219957), gave the strongest statistical evidence in the initial genome-wide study ($P = 1.55 \times 10^{-7}$), and was associated with job-related exhaustion in all the replication sets ($P < 0.05$; $P = 6.75 \times 10^{-7}$ from the meta-analysis). Consistent with studies of mood disorders, individual common genetic variants did not have any strong effect on job-related exhaustion. However, the nominally significant signals from the allelic variant of *UST* in four separate samples suggest that this variant might be a weak risk factor for job-related exhaustion. Together with the previously reported associations of other dermatan/chondroitin sulfate genes with mood disorders, these results indicate a potential molecular pathway for stress-related traits and mark a candidate region for further studies of job-related and general exhaustion.

INTRODUCTION

Job-related exhaustion is the core symptom of burnout, a work-related stress syndrome that consists of three dimensions: exhaustion, cynicism, and diminished professional efficacy. (1). The burnout syndrome is associated with several health problems, including depressive disorders (2), physical illnesses (3), sleep problems (4) and complaints of cognitive impairment (5). The prevalence of severe burnout in the Finnish working population has varied from 2 to 7% (2,6), while mild symptoms have been reported by up to 25% of workers. As severe burnout is linked to an increased risk of medically certified sickness absence, work disability pensions (7), and even all-cause mortality among workers aged under 45 years (8), it imposes an

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economical burden on society in addition to the subjective deleterious effects.

Burnout is not regarded as a disease in the medical classification systems, Classification of diseases and Related Health Problems (ICD-10) or Diagnostic and Statistical Manual of Mental Disorders (DSM-IV), but it correlates moderately with a diagnosis of depression: in the Finnish population, for example, half of the individuals with severe burnout had suffered from major depressive disorder, minor depressive disorder (two to four symptoms) or dysthymia during the previous year. Depressive disorder is also more common in individuals with mild burnout (20.3%) than in those with no symptoms of burnout (7.3%) (2). Job-related exhaustion predicts depressive symptoms 3–4 years later, but not vice versa (9). Importantly, the concepts of burnout and depression are not identical, as both burnout syndrome and job-related exhaustion symptoms are also seen without clinical depression (2). Burnout and, especially, job-related exhaustion both correlate strongly with the personality trait of neuroticism (10). It is possible that burnout is part of the same continuum with internalizing disorders which share part of their genetic background with neuroticism and partly have separate neuroticism-independent genetic risk factors (11).

The heritability of job-related exhaustion was estimated to be 33% for both males and females in a large Swedish study (12), i.e. corresponding to that reported for depression (13). In a smaller Dutch study, the genetic component explained 30% of the variance in males, but only 13% in females (14). To date, the molecular genetic background of job-related exhaustion remains unexplored.

Work stressors, such as excessive work demands, have a fundamental role in the development of burnout (15,16), and job-related exhaustion is based on the concept that the symptoms are a reaction to the work environment. Therefore, we hypothesized that the interaction of genetic vulnerability factors with a stressful work environment is a key element in job-related exhaustion. In order to search for genetic variants contributing to the individual vulnerability to job-related exhaustion we performed a genome-wide association study (GWAS) of 555 388 single-nucleotide polymorphisms (SNPs), using the quantitative exhaustion scale of the Maslach Burnout Inventory-General Survey (MBI-GS) in the GENMETS cohort of the Finnish Health 2000 sample ($n = 1256$) and performed replication analyses for the strongest findings in the rest of the workers in that cohort ($n = 1660$), in two occupational cohorts ($n = 1451$), and in a further Finnish population cohort, FINRISK ($n = 10\,753$) (Fig. 1). To examine the possible interaction of genetic vulnerability and work stress in the etiology of job-related exhaustion, we performed an interaction analysis at the genome-wide level. Given the requirement of large sample sizes for detecting interaction in genetic studies by hypothesis-free analysis, we focused in our gene-environment interaction (G × E) testing mainly on the variants with a main effect on job-related exhaustion.

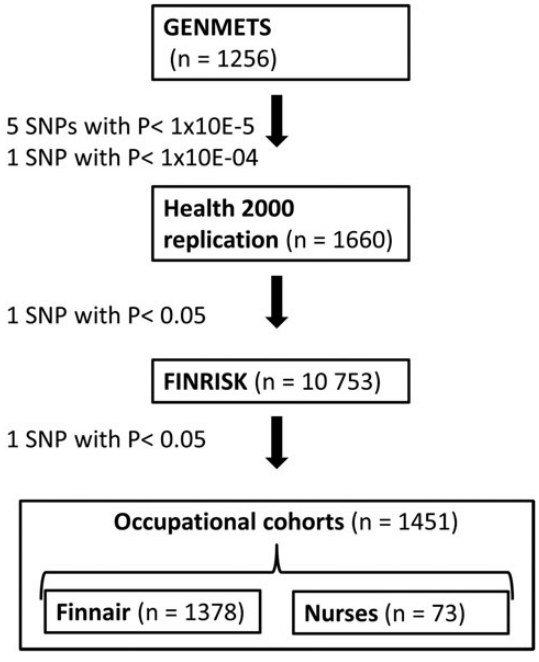

Figure 1. Study flow.

RESULTS

Genome-wide analysis

We performed a genome-wide association analysis for the exhaustion score of MBI-GS in 1256 individuals from the population-based Health 2000 sample. Figure 2A illustrates the quantile–quantile (Q–Q) plot for the observed versus expected $-2\log(P\text{-value})$ and Supplementary Material, Fig. SF1 the Manhattan plot. Some more significant results were seen than expected by chance, but none of these reached the genome-wide significance threshold of $P < 5 \times 10^{-8}$. The complete list of *P*-values is contained in the Supplementary Material (ftp://ftp.techset.co.uk/Sulkava_GWAS/). The most significant association was obtained for rs13219957 ($P = 1.55 \times 10^{-7}$, $\beta = 0.306$, $R^2 = 2.1\%$), which is situated on chromosome 6 in an intron of the gene encoding for uronyl-2-sulfotransferase (*UST*). The results for SNPs with $P < 5 \times 10^{-5}$ for the association in the genome-wide analysis (the level corresponding to the power of 62% when QTL variance was estimated to be 1.5%) are displayed in Table 1. Of the 31 reported SNPs, only two (rs7677237, rs4237913) showed a nominally significant interaction with the subjects' sex ($0.01 < P < 0.05$).

Gene–environment interaction analysis

In order to evaluate our hypothesis of a gene-environment interaction in the etiology of job-related burnout, we used the GENMETS sample to search for evidence of an interaction between work stress as defined by job demands and genetic variants at a genome-wide level, and separately for variants with a suggestive main effect on job-related exhaustion. In the genome-wide interaction analysis, none of the associations were significant ($P < 5 \times 10^{-8}$), and the results did not differ from those expected by chance alone (Supplementary Material, Fig. SF2). SNPs with $P < 5 \times 10^{-5}$ in the interaction analysis are reported in Supplementary Material, Table ST2.

Table 1 shows that 8 of 31 SNPs with a suggestive main effect on job-related exhaustion had a significant interaction with work stress as defined by work demands, and four tests were

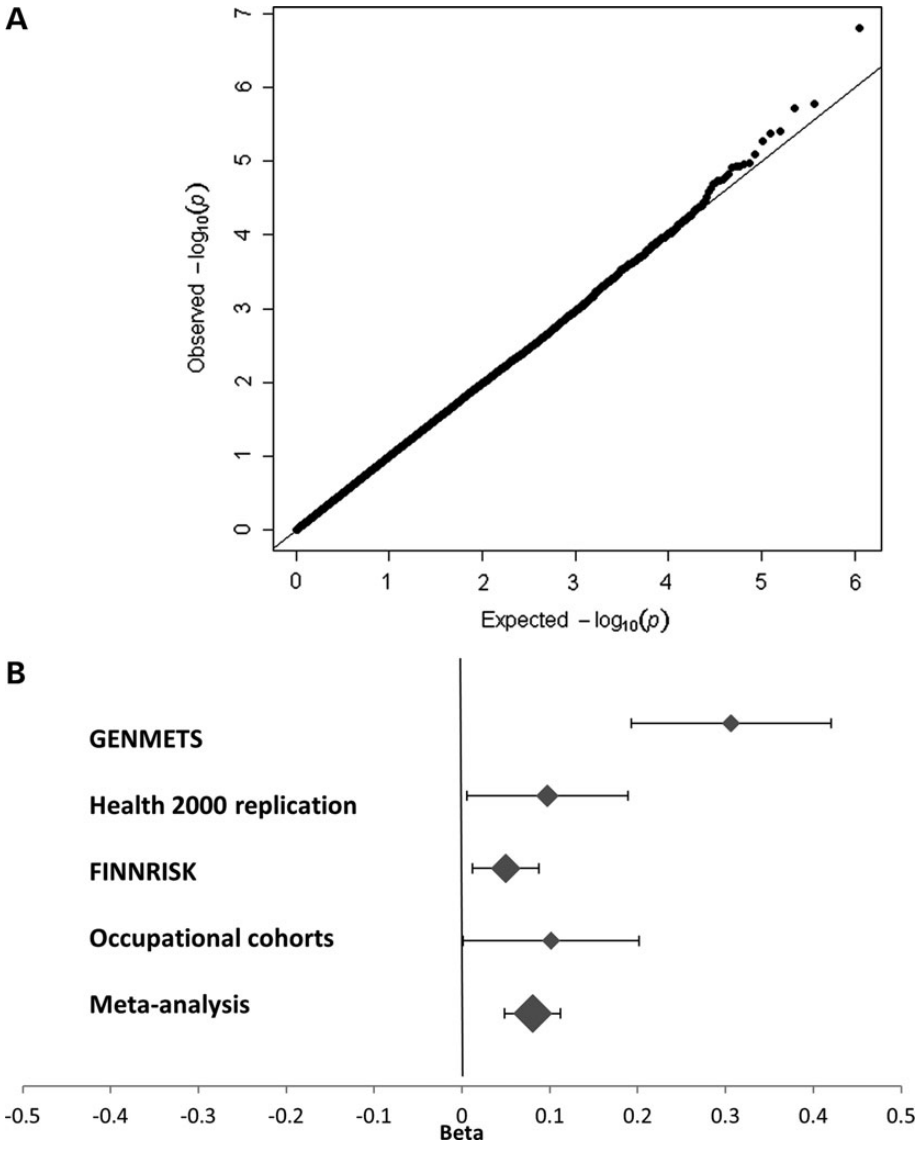

Figure 2. (**A** and **B**) A Q–Q plot of observed versus expected $-\log(P)$ of the GWAS of job-related exhaustion in the GENMETS sample. (B) Effect sizes of rs13219957 in the association analysis with job-related exhaustion in the different cohorts with 95% confidence intervals.

significant after correction for multiple testing (rs10502587, $P_{\text{Bonferroni}} = 0.015$; rs13381088 $P_{\text{Bonferroni}} = 0.006$; rs3786310, $P_{\text{Bonferroni}} = 0.006$; rs3826611, $P_{\text{Bonferroni}} = 0.0062$). These four SNPs were situated in the intron of *FAM59A* and were in a high LD in GENMETS ($D' > 0.8$). The work demand score had a moderate correlation with the exhaustion score (Pearson's correlation $= 0.363$, $P < 0.05$).

Replication analyses and meta-analysis

A replication analysis was performed for five SNPs with a *P*-value of $<1 \times 10^{-5}$ in the rest of the workers ($n = 1660$) from the Health 2000 study. In addition, we selected one SNP with $P < 1 \times 10^{-4}$ (rs10488596 in *CHRM4*, $P_{\text{GWAS}} = 7.01 \times 10^{-5}$) because of its location in the intron of the functionally interesting cholinergic receptor gene. The statistically strongest signal of the genome-wide analysis, from SNP rs13219957 in the *UST* intron, showed also a suggestively significant association with job-related exhaustion in the replication dataset ($P = 0.038$, $\beta = 0.097$) (Table 2), which, though, did not survive correction for multiple testing ($P_{\text{Bonferroni}} = 0.207$). No other variant yielded a suggestive evidence (pointwise $P < 0.05$) in the analysis (Table 2). The gene–environment interaction of rs10502587 in *FAM59A* was not replicated in the Health 2000 replication dataset ($P_{\text{G}\times\text{E}} = 0.782$).

Rs13219957 was genotyped in the Finnish FINRISK population cohort where a one-item measure assessing job-related exhaustion was available for 10 753 workers. We observed an allelic replication for job-related exhaustion at a population level ($P = 0.010$, $\beta = 0.050$). The variant was also associated with the question on the inconvenience caused by time pressure at work ($n = 10\,140$, $P = 0.036$, $\beta = 0.042$) and a question on general exhaustion (not limited to the work context) in the complete FINRISK sample ($n = 20\,813$ $P = 0.049$, $\beta = 0.027$). The

Table 1. Strongest association findings ($P < 5 \times 10^{-5}$) in the genome-wide analysis of job-related exhaustion and interaction with work demands in the GENMETS sample

CHR	SNP	Position (kb)	Gene[a]	Relation to gene	A1	MAF	β	SE	P	β (G×E)	P (G×E)
6	rs13219957	149 270 829	UST	Intronic	A	0.141	0.306	0.058	1.55E − 07	−0.007	0.9191
4	rs7677237	89 306 659	HERC6	Non synonymous coding	G	0.046	0.447	0.093	1.69E − 06	−0.003	0.977
18	rs10502587	29 920 430	FAM59A	Intronic	A	0.102	−0.308	0.064	1.90E − 06	−0.264	0.0005
1	rs11121720	11 364 920	UBIAD1	Intergenic	G	0.061	0.382	0.082	3.91E − 06	0.239	0.0127
15	rs2034705	36 832 625	C15orf41	Intergenic	G	0.153	−0.252	0.055	4.20E − 06	−0.010	0.8698
1	rs17036631	11 347 492	UBIAD1	Untranslated-3	A	0.062	0.371	0.081	5.41E − 06	0.242	0.0102
2	rs2420382	60 896 740	PAPOLG	Intergenic	C	0.211	0.227	0.051	8.15E − 06	0.104	0.0604
12	rs1696422	130 568 190		Intergenic	A	0.303	−0.191	0.043	1.05E − 05	0.014	0.7678
9	rs7870439	100 951 838	CORO2A	Intronic	A	0.210	0.216	0.049	1.10E − 05	0.051	0.342
12	rs901964	48 676 038		Intergenic	A	0.286	0.194	0.044	1.20E − 05	−0.075	0.1311
12	rs1489111	48 679 615		Intergenic	A	0.286	0.194	0.044	1.20E − 05	−0.075	0.1311
12	rs2634685	48 768 555		Intergenic	G	0.325	0.188	0.043	1.21E − 05	−0.063	0.1906
12	rs10783231	48 578 325	c12orf68	Non-synonymous coding	A	0.289	0.191	0.044	1.48E − 05	−0.073	0.1408
12	rs826917	130 567 562	FZD10	Intergenic	G	0.213	−0.207	0.048	1.65E − 05	0.021	0.718
8	rs7018287	134 264 054	NDRG1	Intronic	A	0.118	0.274	0.064	1.78E − 05	0.116	0.0891
12	rs4237913	96 162 288	NTN4	Intronic	G	0.180	0.226	0.053	1.84E − 05	−0.004	0.9493
15	rs2467350	36 772 809		Intergenic	C	0.373	−0.175	0.041	1.87E − 05	0.009	0.8294
15	rs2444751	36 777 549		Intergenic	A	0.373	−0.174	0.041	1.96E − 05	0.010	0.8274
9	rs3780452	100 964 701	TBC1D2	Intronic	A	0.488	−0.168	0.039	2.03E − 05	−0.014	0.7526
9	rs3780453	100 964 651	TBC1D2	Intronic	A	0.498	0.166	0.039	2.37E − 05	0.008	0.8542
12	rs7316408	118 417 779	KSR2	Intergenic	G	0.368	0.181	0.043	2.60E − 05	0.022	0.6464
4	rs12649785	37 335 699	KIAA1239	Intronic	C	0.127	0.252	0.060	3.04E − 05	−0.096	0.151
6	rs11153162	97 204 294	GPR63	Downstream	A	0.245	0.194	0.047	3.48E − 05	0.080	0.1208
18	rs13381088	29 969 986	FAM59A	Intronic	A	0.115	−0.258	0.062	3.64E − 05	−0.266	0.0002
18	rs3786310	29 972 724	FAM59A	Intronic	G	0.115	−0.256	0.062	4.05E − 05	−0.266	0.0002
6	rs2744604	24 540 539	ALDH5A1	Intergenic	A	0.325	−0.179	0.044	4.21E − 05	−0.097	0.0417
3	rs9850123	192 042 183	FGF12	Intronic	G	0.145	0.242	0.059	4.23E − 05	0.009	0.8867
8	rs11135876	25 477 382		Intergenic	A	0.238	−0.199	0.049	4.47E − 05	−0.030	0.5578
2	rs17031423	60 893 016		Intergenic	G	0.221	0.204	0.050	4.49E − 05	0.117	0.0322
18	rs3826611	29 972 565	FAM59A	Intronic	G	0.115	−0.253	0.062	4.72E − 05	−0.265	0.0002
3	rs2886242	191 189 965	PYDC2	Intergenic	G	0.156	0.236	0.058	4.91E − 05	−0.004	0.9492

SNP, single-nucleotide polymorphism; CHR, chromosome; kb, kilobase; A1, minor allele; MAF, minor allele frequency; G×E, gene–environment interaction. Nominal P-values are reported.
[a]The nearest gene is reported if there is one closer than 100 kb from the marker.

effect on general exhaustion was slightly stronger among workers ($n = 12\,812$, $P = 0.085$, $\beta = 0.029$) than non-workers ($n = 8001$, $P = 0.394$, $\beta = 0.020$), even though the association in workers did not reach the nominal level of significance.

The effect of rs13219957 on job-related exhaustion was further tested in two occupational cohorts: a cohort of employees of an airline company ($n = 1378$) and a cohort of nurses ($n = 73$). These two samples were combined for the analysis because of the small sample size in the nurse cohort. Their meta-analysis showed a significant association of rs13219957 with the MBI-GS exhaustion score ($P = 0.047$, $\beta = 0.102$)

Finally, we performed a fixed-effect meta-analysis across all the study samples (total $n = 15\,120$), which yielded a P-value of 6.75×10^{-7} and a normalized β-value of 0.081. The results of the replication studies and the meta-analysis for rs13219957 are reported in Table 3.

Association of rs13219957 with the different domains of burnout symptoms and with depressive symptoms

The Burnout syndrome consists of three dimensions: exhaustion, cynicism and diminished professional efficacy, which are reflected by three subscales of MBI-GS. We calculated the associations of rs13219957 with other symptoms of the burnout syndrome in the cohorts which had primarily been analysed for job-related exhaustion. In the Health 2000 population samples, the strongest association was seen with the MBI's exhaustion subscale (GENMETS: exhaustion $P = 1.55 \times 10^{-7}$, $\beta = 0.306$, cynicism $P = 0.005$, $\beta = 0.165$, diminished professional efficacy $P = 0.014$, $\beta = 0.143$) (Table 4). In the occupational cohorts combined, the signal was stronger for diminished professional efficacy ($P = 0.025$, $\beta = 0.102$) when compared with exhaustion ($P = 0.047$, $\beta = 0.102$), but in a separate analysis of the nurse cohort, the signal was strongest for cynicism ($P = 0.015$, $\beta = 0.513$), possibly because of the specific natures of the occupational groups.

Considering the co-morbid pattern of job-related exhaustion and depression, and the risk of job-related exhaustion on subsequent depressive disorder, we also assessed the association of rs1329957 with symptoms of depression, including anhedonia, somatic symptoms and appetite. Similarly weighted principal components were found in every cohort, even though the measures for depressive symptoms slightly varied between them (see Materials and methods). No significant association of rs13219957 with depressive symptoms was seen in the Health 2000 replication sample or in the FINNRISK sample ($P > 0.05$). In the Finnair sample, a significant association was observed only with the somatic symptoms of depression ($P = 0.014$, $\beta = 0.129$). In the GENMETS dataset, a less significant signal compared with the job-related exhaustion signal was

Table 2. Results of the replication analysis of job-related exhaustion in the Health 2000 replication sample and the pooled Health 2000 sample

SNP	CHR	Gene	A1	MAF	β_{repl}	SE_{repl}	P_{repl}	β_{comb}	SE_{comb}	P_{comb}
rs13219957	6	UST	A	0.150	0.097	0.047	0.038	0.178	0.036	9.68E − 07
rs7677237	4	HERC6	C	0.040	0.042	0.088	0.633	0.237	0.063	0.0002
rs10502587	18	FAM59A	T	0.105	−0.007	0.054	0.892	−0.134	0.041	0.0013
rs11121720	1	UBIAD1	C	0.064	0.057	0.070	0.417	0.196	0.053	0.0002
rs2420382	2	PAPOLG	C	0.218	0.003	0.042	0.944	0.095	0.032	0.0033
rs10488596	7	CHRM2	T	0.152	0.018	0.047	0.701	0.110	0.036	0.0021

SNP, single-nucleotide polymorphism; CHR, chromosome; A1, minor allele; β_{repl}, beta for the analysis of the replication dataset; P_{repl}, *P*-value for the analysis of the replication dataset; β_{comb}, β for the analysis of the combined dataset; SE_{comb}, SE for the analysis of the combined Health 2000 dataset; P_{comb}, *P*-value for the analysis of the combined Health 2000 dataset.
Nominal *P*-values are reported. Of the seven SNPs with $P < 1 \times 10^{-5}$ in GENMETS two were not included in the replication analysis: rs17036631 because of the strong linkage ($D' = 1.00$) with rs11121720, and rs2034705 due to technical problems in genotyping.

Table 3. Association of rs13219957 with job-related exhaustion in the Health 2000 samples, the FINRISK sample, and the occupational samples, and the meta-analysis of the studies

	Sample	*P*	β	SE	MAF	*n*
Health 2000	GENMETS	1.55E − 07	0.306	0.058	0.139	1256
	Health 2000 replication	0.037	0.097	0.047	0.150	1660
FINRISK	FINRISK	0.010	0.050	0.019	0.143	10 753
Occupational cohorts	Finnair	0.093	0.089	0.053	0.152	1378
	Nurses	0.151	0.309	0.213	0.169	73
	Occupational cohorts meta-analysis	0.047	0.102	0.051	0.153	1451
All	Meta-analysis[a,b]	6.75E − 07	0.081	0.016	0.144	15 120

MAF, minor allele frequency.
Nominal *P*-values are reported.
[a]Health 2000 GWAS and the replication sample are pooled together and combined with the other cohorts, using meta-analysis.
[b]*Q* test *P*-value = 0.012, $I^2 = 73\%$, suggesting some heterogeneity which is mostly caused by the FINRISK sample with its different measures of job-related exhaustion.

detected (Table 4). In the combined Health 2000 sample, a suggestive association with Beck Depression Inventory (BDI) components disappeared ($P > 0.05$) after adjustment for the job-related exhaustion score, whereas the association with job-related exhaustion was only marginally diminished ($P < 0.05$) when adjusting for BDI components (Supplementary Material, Tables ST3 and ST4).

DISCUSSION

To our knowledge, this is the first study to examine the genetic basis of job-related exhaustion with molecular genetic methods. We performed the GWAS for the quantitative exhaustion dimension of burnout in a sample of 1256 individuals. The strongest association found, rs13219957 ($P = 1.55 \times 10^{-7}$, $\beta = 0.306$) in the intron of *UST*, was replicated at the $P < 0.05$ level in the replication sample from the same study and in two occupational cohorts. The genetic variant was also associated with a single question concerning job-related exhaustion in a further population-based sample of 10 753 individuals ($P = 0.01$, $\beta = 0.050$). Finally, a meta-analysis including all 15 120 Finnish individuals yielded a genome-wide suggestively significant association ($P = 6.75 \times 10^{-7}$, $\beta = 0.081$). Even though there is a lack of strict genome-wide significance, nominally significant replications in three replication samples suggest that our results are unlikely to be a false-positive finding. Rs13219957, which is situated in the intronic region of the *UST*, marks a candidate region for further studies of job-related and general exhaustion.

Our results show no risk variants of a large or intermediate effect size and genome-wide significance in the association analysis, which is consistent with previous genome-wide studies and a meta-analysis on depression (17,18). However, a surplus of low detected *P*-values compared with expected ones (Fig. 2A) suggests the presence of variants with weak genetic effects that cannot be detected with adequate significance levels by the GWAS with our sample size. This is further supported by the replication of the association of the GWAS top finding, rs13219957, at $P < 0.05$, when using large sufficient population samples.

The marker rs13219957 is situated in the intronic region of the *UST* (uronyl-2-sulfotransferase) gene. Uronyl-2-sulfotransferase is an enzyme that catalyzes the transfer of a sulfate group to dermatan or chondroitin sulfate (19). Dermatan and chondroitin sulfate form the polysaccharide part of chondroitin sulfate proteoglycans (CSPGs), which are an abundant subtype of proteoglycans in the extracellular matrix of nervous system (20). Highly negative sulfate groups of proteoglycans have been proposed to play a part in the binding of growth factors (21) and also to enhance the Alzheimer-like changes in tau protein (22) and amyloid β peptide (23). The importance of CSPGs and chondroitin/dermatan sulfate sulfotransferases for the brain has been illustrated by findings that show their high expression in neurogenic regions of the central nervous system mice, while an inhibition of sulfation was demonstrated to decrease the proliferation of cultured neural stem cells (24). Mice with inhibited *UST* showed a

Table 4. Association of rs13219957 with symptoms of burnout and depression in all the cohorts

	Burnout syndrome Job-related exhaustion		Cynisism		Diminished professional efficacy		Depression Anhedonia		Somatic symptoms		Appetite	
	P	β	*P*	β	*P*	β	*P*	β	*P*	β	*P*	β
GENMETS	**1.55E − 07**	0.306	**0.005**	0.165	**0.014**	0.143	**0.004**	0.127	0.004	0.117	**0.009**	0.107
Health 2000 replication	**0.037**	0.097	0.118	0.073	0.508	0.031	0.856	0.006	0.861	0.005	0.620	0.016
FINNRISK	**0.010**	0.050	NA	NA	NA	NA	0.426	0.025	0.713	0.011	0.227	0.040
Finnair	0.093	0.089	0.257	0.054	**0.032**	0.100	0.216	0.065	**0.014**	0.129	NA	NA
Nurses	0.151	0.309	**0.015**	0.513	0.520	0.137	NA	NA	NA	NA	NA	NA
Occupational cohorts meta-analysis	**0.047**	0.102	0.096	0.078	**0.025**	0.102	NA	NA	NA	NA	NA	NA

Nominal *P*-values are reported.
P-values <0.05 are shown in boldface.

disturbed neuronal migration during brain development (25), which could also affect the functioning of the mature brain.

In a twin study (14), genetic factors explained most of the correlation of job-related exhaustion and anxious depression. In our study, the association of rs13219957 with depressive symptoms was significant in the GENMETS dataset and one of the replication cohorts, the Finnair sample, in which it was associated with the somatic symptoms component of depression (Table 4). The component weights questions of fatigue and loss of energy which are also key elements in the burnout syndrome. This can be interpreted as a certain specificity of the rs13219957 for exhaustion-like symptoms that can be assessed by job-related exhaustion measures, but also by measures that weight somatic symptoms of depression among workers. Homogenization of the sample when restricting the analysis to workers only in the study of job-related exhaustion can affect opportunities to detect an association. More studies are needed to reveal the genetic overlap of job-related exhaustion and depression and to determine whether the genetic factors differ when high work stress is present.

The association of the UST variant with depression symptoms was in this study not as strong as with exhaustion symptoms. However, an intronic variant of *UST* (rs2500535) has been reported to be associated with antidepressive treatment response at a genome-wide significance level (26) (not in linkage disequilibrium with rs13219957 in the Finnish population). In addition, *UST* shows a functional connection with the genes presented in the GWASs of depression. In one study (27), the strongest signal for association ($P = 1.8 \times 10^{-7}$) was observed for an SNP located in the vicinity of *DSEL* (dermatan-sulfate epimerase-like), which, like *UST*, is considered to be involved in the posttranslational modification of dermatan sulfate (28). However, in a meta-analysis of depression, there was no further support for this result (29). In several earlier studies (30–33), the genomic region containing *DSEL* has also been found to be linked to bipolar mood disorder. The close functional similarity of *UST* and *DSEL* may be an indication of the common biological mechanisms for job-related exhaustion and mood disorders. Further support for the role of the dermatan sulfate pathway in the regulation of mood was obtained in a recent meta-analysis of major depressive disorder (34), where a variant of the dermatan chondroitin/sulfate proteoglycan gene, VCAN, emerged as the second-strongest finding.

The gene-environment-wide analysis did not reveal an interaction between job demands and selected genetic variants, but considering the large sample sizes needed for studying G × E (35), it is quite likely that we were underpowered to detect such phenomena. Gene–environment analysis of the strongest findings of the original GWAS did not show any replicable interaction. Interestingly, when using samples derived from workers of an airline company or nurses rather than those from the general population, rs13219957 showed stronger associations with diminished professional efficacy and cynicism symptoms than with job-related exhaustion. This could reflect the combined effect of the genetic risk factors and the work environment on the manifestation of burnout. The G × E part of our study must be seen as an initial explorative trial to open the discussion on the topic.

The moderate size of the original sample in the GWAS is a limitation of our study, which was partially balanced by the use of substantially larger replication samples. This did enable us to confirm the association of rs13219957 with job-related exhaustion, but the moderate original sample size decreased the power, increased the rate of type II errors and made it very likely that we did not detect all of the common genetic variation linked to job-related exhaustion (Power calculation, Supplementary Material). Job-related exhaustion was evaluated with a subscale of a burnout questionnaire (MBI-GS), which is a standard procedure to assess burnout (36), because there are at present no biological markers or other validated objective measures of burnout available. In the FINRISK sample, the job-related exhaustion was evaluated with a single question concerning workers' experience regarding high work load. A single unvalidated question is not equivalent to the exhaustion subscale in the MBI-GS questionnaire, which can be the reason for the lowest effect size seen in this cohort and for the heterogeneity in the meta-analysis when the FINRISK cohort is included (Table 3). However, the effect noticed in relation to also the other work-stress-related question suggests that the question indeed relates to the job-related exhaustion phenomenon (Results). In the G × E study, the measurement of job demands is likely to be affected by the mental state of the responders causing a common method problem with job-related exhaustion questions.

In sum, we report here the first attempt to explore the genetic background of job-related exhaustion. The variant of *UST* with the strongest association in our genome-wide analysis showed

a significant association also in three replication samples. The results give quite strong support for the idea that the variant marks an area with a weak regulating effect on such exhaustion symptoms that are seen in the context of professional work. Fine mapping of the genomic area and functional studies are still needed to locate the effective variant. Association studies in populations outside Finland are needed to assess how our findings can be generalized. Considering previous reports about associations of the other dermatan/chondroitin sulfate-related genes with mood disorders, the whole molecular pathway should be regarded as a candidate pathway for studies of stress-related traits. The hypothesis that there are interactions between genes and stressful work environment in job-related exhaustion must be examined further using longitudinal study settings and larger samples.

MATERIALS AND METHODS

Population and sampling

The subjects for this study were obtained from two Finnish population-based studies and two Finnish occupational samples: the Health 2000 study including the GENMETS GWAS dataset and the replication dataset, the FINRISK cohorts, the Finnair airline company sample and the nurse sample.

Health 2000 is a large epidemiological health study carried out in 2000–2001 (http://www.terveys2000.fi/doc/methodologyrep.pdf). The sample collection was performed in two stages with stratified cluster sampling, and it was representative of the Finnish population aged 30 years and over. The study design has been described in detail earlier (37). Characteristics of the sample for this genetic study are shown in Supplementary Material, Table ST1. The Ethics Committee of the Helsinki and Uusimaa Hospital District approved the study protocol, and a written informed consent was obtained from all participants after providing a description of the study.

The Health 2000 sample comprised 8028 adults aged 30 years and over; 93% had participated in at least some phase of the study. Inclusion criteria for our genetic study comprised an age of $\leq$64 years, currently working or on sick leave, and no more than one missing value per dimension of the MBI-GS. These criteria were met by 3441 individuals, and DNA samples had been obtained from 3253 of them. Of the 3253 individuals, 1456 were genotyped at a genome-wide level in the context of the GENMETS substudy of the Health 2000 survey. The GENMETS sample comprised of the metabolic syndrome cases and the controls (38). Selection criteria for the GENMETS study are shown in the Supplementary Material. To minimize the selection effect, the case–control status in the GENMETS study was used as a covariate in the genetic analyses. Those 1797 Health 2000 individuals who satisfied the phenotypic inclusion criteria of the present study, but were not included in GENMETS, were used as a replication sample for the strongest findings from the genome-wide study.

FINRISK is a Finnish population study of non-communicable disorders in which a new cohort is collected every 5 years (39). The population sampling of the FINRISK cohorts is described in detail in the Supplementary Material. In this study, we used three FINRISK samples, from 1997, 2002 and 2007; a single question related to job-related exhaustion was included in 1997 and 2002, questionnaires and a question on general exhaustion in all of the years. A written informed consent was obtained from all participants. The appropriate ethics committees approved the surveys.

The FINRISK cohorts in 1997, 2002 and 2007 comprised, together, 32 549 individuals (1997: $n = 7159$, response rate = 71.6%; 2002: $n = 13\,437$, response rate = 71.3%, 2007: $n = 11\,953$, response rate = 66.9%). An answer to the question about job-related exhaustion was available for 12 493 workers, and about general exhaustion for 25 424 subjects, both workers and non-workers.

The Finnair airline company sample included workers who were occupied in different ground service functions at the Helsinki Vantaa airport, or as pilots or cabin attendants (40). The nurse sample was derived from a bigger study of female health care professionals, which was described in detail earlier (41). All of the nurses were working in shifts and whether in high stress or low stress environments. Altogether, we had for 1518 employees of the Finnair airline company and for 77 nurses information from MBI-GS. The occupational samples were analysed together using fixed-effect meta-analysis.

Measures

Job-related exhaustion was assessed in the Health 2000 study and in the occupational cohorts using the exhaustion subscale of the Finnish version of the MBI-GS (42). MBI-GS is a widely used questionnaire that covers three dimensions of burnout with separate subscales (43). The method is applicable to any profession, and its reliability and validity have been confirmed in different samples (44,45). All the subscales consist of five symptom statements scored on a seven-item frequency scale ranging from 0 (never) to 6 (daily). One missing value per subscale was accepted and replaced with a mean value.

In the FINRISK sample, job-related exhaustion was assessed with a single question: how often are you troubled by having to stretch your strength to the extreme to be able to cope with your present work or work load? The five-stage response scale ranged from almost always to never. Only workers responded to the question. In addition we asked: how often are you troubled by continuous busyness/stress at your work? This question was similarly scaled. General exhaustion was measured by the simple question assessing feelings during the past month: 'Do you feel exhausted or overworked?'. The response scale ranged from 1 to 3 (1 = often, 2 = sometimes, 3 = not at all). All the scales were inversed for the analyses to make the comparison with other results easier. These variables followed the normal distribution.

In secondary analyses of Health 2000 and the occupational cohorts, we used two other subscales of MBI-GS, which included five questions concerning cynicism and six concerning professional efficacy. Also the best measure of depressive symptoms was selected for each cohort. Three principal components from the 21-item BDI (46) were formed for the Health 2000 sample, using the principal component analysis (PCA). The first component focused on anhedonia, the second on somatic symptoms and the third on appetite (Supplementary Material, Tables ST6 and ST7). To achieve a normal distribution of the principal components of BDI, we used the $1/\times$transformation of the principal components for

anhedonia and appetite and the log transformation of the principal component for somatic symptoms. In the FINRISK sample, the 13-item BDI was factorized with PCA. Three components corresponding to the components of the 21-item BDI were detected (Supplementary Material, Tables ST7 and ST8). Also in this sample, the $1/\times$ transformation was made for the anhedonia and appetite components. In the Finnair cohort, PCA was made for the 10-item depression scale (DEPS) (47), which yielded two components corresponding to the andhedonia and somatic symptoms components of BDI (Supplementary Material, Tables ST9 and ST10). Logarithmic transformation was performed on the anhedonia component. Independent of the transformation, all the scores are reported so that higher score means worse symptoms.

The level of demands at work was used as an indicator of the environment's stressfulness. It was evaluated by using a mean score for the five questions in the Job Content Questionnaire (48). The demand score was normally distributed in the sample.

DNA extraction

DNA was extracted from peripheral blood leukocytes using the commercially available Puregene DNA isolation kit (Puregene, Gentra Systems, Minneapolis, MN, USA). The extraction method is described in more detail by Blin and Stafford (49).

Genotyping and quality control

The GENMETS samples were genotyped with the Illumina 610K platform (Illumina Inc., San Diego, CA, USA) at the Wellcome Trust Sanger Institute (Hinxton, UK). The call rate was $>95\%$ for both individuals and markers. Markers with a minor allele frequency (MAF) $<1\%$ or a Hardy–Weinberg $P < 1 \times 10^{-6}$ were excluded from the analyses. Heterozygosity and gender checks were performed, and individuals with any discrepancies were removed. Cryptic relatedness was detected for six individuals, and also they were removed. The genotyping was successful for 1256 individuals with job-related exhaustion information and 555 388 SNPs. In order to control for population stratification in GENMETS, 20 principal components were created and used as covariates in the genome-wide analysis. The replication sets were genotyped at the Institute for Molecular Medicine Finland using iPLEX Gold chemistry (Sequenom, San Diego, CA, USA). In the initial health 2000 replication set, for all six SNPs, the success rate was $>95\%$, the MAF >0.01 and the Hardy–Weinberg equilibrium, $P > 0.001$. The call rate for genotyping of individuals was 95%, and this was passed by 1660 individuals with a non-missing phenotype. The same quality standards for genotyping were required in the occupational cohorts.

Rs13219957 was genotyped in the FINRISK 1997, 2002 and 2007 cohorts using iPLEX Gold chemistry (Sequenom, San Diego, CA, USA). Individuals with a call rate exceeding 80% for the iplex were kept in the analysis ($n = 21\,390$). Genotyping of rs13219957 was successful for 21 256 individuals (99.4%), 10 753 of whom had answered to the question concerning job-related exhaustion and 20 813 to the question about general exhaustion. Genotyping was performed at the Institute for Molecular Medicine Finland. The same genotyping technique was performed in the occupational cohorts and genotyping was successful for 73 individuals from the nurse cohort and 1378 individuals from the Finnair cohort.

Statistical analysis

The exhaustion score was logarithmically transformed in order to follow normal distribution and then treated as a quantitative trait. The association between genotyped SNPs and the exhaustion score was tested using the linear regression analysis of the PLINK software. We expected the additive model to be valid, meaning that one copy of an allele would have less of an effect than two copies, but more of an effect than no copies. *Z*-score normalization was performed for all the analyses, meaning that distribution was standardized for a mean of 0 and a SD of 1. Beta (β) stands in this article for the effect of one minor allele in units of standard deviation. Age, sex, principal components for GWAS data, and metabolic syndrome status for GENMETS were used as covariates in the GWAS. Linear regression analysis with age and sex as covariates was performed in the rest of the analyses. R^2 stands for a coefficient of determination, indicating how much the genetic variant explains about the variance in job-related exhaustion after removal of the variance caused by age and sex. The Bonferroni correction for the Health 2000 replication sample was calculated according to the equation $P_{\text{Bonferroni}} = 1-(1 - \text{P})^{\text{number of tests}}$. The programs used in this article are listed in detail in the Supplementary Material.

SUPPLEMENTARY MATERIAL

Supplementary Material is available at *HMG* online.

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Conflict of Interest statement. All authors report no conflicts of interest.

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REFERENCES

1. Maslach, C., Schaufeli, W.B. and Leiter, M.P. (2001) Job burnout. *Annu. Rev. Psychol.*, **52**, 397–422.
2. Ahola, K., Honkonen, T., Isometsa, E., Kalimo, R., Nykyri, E., Aromaa, A. and Lonnqvist, J. (2005) The relationship between job-related burnout and depressive disorders—results from the Finnish health 2000 study. *J. Affect. Disord.*, **88**, 55–62.
3. Honkonen, T., Ahola, K., Pertovaara, M., Isometsa, E., Kalimo, R., Nykyri, E., Aromaa, A. and Lonnqvist, J. (2006) The association between burnout and physical illness in the general population—results from the Finnish health 2000 study. *J. Psychosom. Res.*, **61**, 59–66.
4. Ekstedt, M., Soderstrom, M., Akerstedt, T., Nilsson, J., Sondergaard, H.P. and Aleksander, P. (2006) Disturbed sleep and fatigue in occupational burnout. *Scand. J. Work Environ. Health*, **32**, 121–131.
5. Sandstrom, A., Rhodin, I.N., Lundberg, M., Olsson, T. and Nyberg, L. (2005) Impaired cognitive performance in patients with chronic burnout syndrome. *Biol. Psychol.*, **69**, 271–279.
6. Kalimo, R. (2000) The challenge of changing work and stress for human resources. The case of finland. *J. Tokyo Med. Univ.*, **58**, 349–356.
7. Ahola, K., Gould, R., Virtanen, M., Honkonen, T., Aromaa, A. and Lonnqvist, J. (2009) Occupational burnout as a predictor of disability pension: a population-based cohort study. *Occup. Environ. Med.*, **66**, 284–290, discussion 282–283.
8. Ahola, K., Vaananen, A., Koskinen, A., Kouvonen, A. and Shirom, A. (2010) Burnout as a predictor of all-cause mortality among industrial employees: a 10-year prospective register-linkage study. *J. Psychosom. Res.*, **69**, 51–57.
9. Hakanen, J.J. and Schaufeli, W.B. (2012) Do burnout and work engagement predict depressive symptoms and life satisfaction? A three-wave seven-year prospective study. *J. Affect. Disord.*, **141**, 415–424.
10. Swider, B.W. and Zimmerman, R.D. (2010) Born to burnout: a meta-analytic path model of personality, job burnout, and work outcomes. *J. Vocat. Behav.*, **76**, 487–506.
11. Hettema, J.M., Neale, M.C., Myers, J.M., Prescott, C.A. and Kendler, K.S. (2006) A population-based twin study of the relationship between neuroticism and internalizing disorders. *Am. J. Psychiatry*, **163**, 857–864.
12. Blom, V., Bergstrom, G., Hallsten, L., Bodin, L. and Svedberg, P. (2012) Genetic susceptibility to burnout in a Swedish twin cohort. *Eur. J. Epidemiol.*, **27**, 225–231.
13. Sullivan, P.F., Neale, M.C. and Kendler, K.S. (2000) Genetic epidemiology of major depression: review and meta-analysis. *Am. J. Psychiatry*, **157**, 1552–1562.
14. Middeldorp, C.M., Cath, D.C. and Boomsma, D.I. (2006) A twin-family study of the association between employment, burnout and anxious depression. *J. Affect. Disord.*, **90**, 163–169.
15. Lindblom, K.M., Linton, S.J., Fedeli, C. and Bryngelsson, I.L. (2006) Burnout in the working population: relations to psychosocial work factors. *Int. J. Behav. Med.*, **13**, 51–59.
16. Magnusson Hanson, L.L., Theorell, T., Oxenstierna, G., Hyde, M. and Westerlund, H. (2008) Demand, control and social climate as predictors of emotional exhaustion symptoms in working Swedish men and women. *Scand. J. Public Health*, **36**, 737–743.
17. Wray, N.R., Pergadia, M.L., Blackwood, D.H., Penninx, B.W., Gordon, S.D., Nyholt, D.R., Ripke, S., Macintyre, D.J., McGhee, K.A., Maclean, A.W. *et al.* (2010) Genome-wide association study of major depressive disorder: new results, meta-analysis, and lessons learned. *Mol. Psychiatry*, **17**, 36–48.
18. Major Depressive Disorder Working Group of the Psychiatric GWAS Consortium (2012). A mega-analysis of genome-wide association studies for major depressive disorder. *Mol. Psychiatry*, **18**, 497–511.
19. Kobayashi, M., Sugumaran, G., Liu, J., Shworak, N.W., Silbert, J.E. and Rosenberg, R.D. (1999) Molecular cloning and characterization of a human uronyl 2-sulfotransferase that sulfates iduronyl and glucuronyl residues in dermatan/chondroitin sulfate. *J. Biol. Chem.*, **274**, 10474–10480.
20. Bovolenta, P. and Fernaud-Espinosa, I. (2000) Nervous system proteoglycans as modulators of neurite outgrowth. *Prog. Neurobiol.*, **61**, 113–132.
21. Pan, J., Qian, Y., Zhou, X., Lu, H., Ramacciotti, E. and Zhang, L. (2010) Chemically oversulfated glycosaminoglycans are potent modulators of contact system activation and different cell signaling pathways. *J. Biol. Chem.*, **285**, 22966–22975.
22. Hasegawa, M., Crowther, R.A., Jakes, R. and Goedert, M. (1997) Alzheimer-like changes in microtubule-associated protein tau induced by sulfated glycosaminoglycans. Inhibition of microtubule binding, stimulation of phosphorylation, and filament assembly depend on the degree of sulfation. *J. Biol. Chem.*, **272**, 33118–33124.
23. Castillo, G.M., Lukito, W., Wight, T.N. and Snow, A.D. (1999) The sulfate moieties of glycosaminoglycans are critical for the enhancement of beta-amyloid protein fibril formation. *J. Neurochem.*, **72**, 1681–1687.
24. Akita, K., Von Holst, A., Furukawa, Y., Mikami, T., Sugahara, K. and Faissner, A. (2008) Expression of multiple chondroitin/dermatan sulfotransferases in the neurogenic regions of the embryonic and adult central nervous system implies that complex chondroitin sulfates have a role in neural stem cell maintenance. *Stem Cells*, **26**, 798–809.
25. Ishii, M. and Maeda, N. (2008) Oversulfated chondroitin sulfate plays critical roles in the neuronal migration in the cerebral cortex. *J. Biol. Chem.*, **283**, 32610–32620.
26. Uher, R., Perroud, N., Ng, M.Y., Hauser, J., Henigsberg, N., Maier, W., Mors, O., Placentino, A., Rietschel, M., Souery, D. *et al.* (2010) Genome-wide pharmacogenetics of antidepressant response in the GENDEP project. *Am. J. Psychiatry*, **167**, 555–564.
27. Shi, J., Potash, J.B., Knowles, J.A., Weissman, M.M., Coryell, W., Scheftner, W.A., Lawson, W.B., DePaulo, J.R. Jr, Gejman, P.V., Sanders, A.R. *et al.* (2011) Genome-wide association study of recurrent early-onset major depressive disorder. *Mol. Psychiatry*, **16**, 193–201.
28. Maccarana, M., Olander, B., Malmstrom, J., Tiedemann, K., Aebersold, R., Lindahl, U., Li, J.P. and Malmstrom, A. (2006) Biosynthesis of dermatan sulfate: Chondroitin-glucuronate C5-epimerase is identical to SART2. *J. Biol. Chem.*, **281**, 11560–11568.
29. Shyn, S.I., Shi, J., Kraft, J.B., Potash, J.B., Knowles, J.A., Weissman, M.M., Garriock, H.A., Yokoyama, J.S., McGrath, P.J., Peters, E.J. *et al.* (2011) Novel loci for major depression identified by genome-wide association study of sequenced treatment alternatives to relieve depression and meta-analysis of three studies. *Mol. Psychiatry*, **16**, 202–215.
30. Goossens, D., Van Gestel, S., Claes, S., De Rijk, P., Souery, D., Massat, I., Van den Bossche, D., Backhovens, H., Mendlewicz, J., Van Broeckhoven, C. *et al.* (2003) A novel CpG-associated brain-expressed candidate gene for chromosome 18q-linked bipolar disorder. *Mol. Psychiatry*, **8**, 83–89.
31. Verheyen, G.R., Villafuerte, S.M., Del-Favero, J., Souery, D., Mendlewicz, J., Van Broeckhoven, C. and Raeymaekers, P. (1999) Genetic refinement and physical mapping of a chromosome 18q candidate region for bipolar disorder. *Eur. J. Hum. Genet.*, **7**, 427–434.
32. Nwulia, E.A., Miao, K., Zandi, P.P., Mackinnon, D.F., Depaulo, J.R. Jr. and Mcinnis, M.G. (2007) Genome-wide scan of bipolar II disorder. *Bipolar Disord.*, **9**, 580–588.
33. Fallin, M.D., Lasseter, V.K., Wolyniec, P.S., McGrath, J.A., Nestadt, G., Valle, D., Liang, K.-. and Pulver, A.E. (2004) Genomewide linkage scan for bipolar-disorder susceptibility loci among Ashkenazi jewish families. *Am. J. Hum. Genet.*, **75**, 204–219.
34. Lewis, C.M., Ng, M.Y., Butler, A.W., Cohen-Woods, S., Uher, R., Pirlo, K., Weale, M.E., Schosser, A., Paredes, U.M., Rivera, M. *et al.* (2010) Genome-wide association study of major recurrent depression in the UK population. *Am. J. Psychiatry*, **167**, 949–957.
35. Duncan, L.E. and Keller, M.C. (2011) A critical review of the first 10 years of candidate gene-by-environment interaction research in psychiatry. *Am. J. Psychiatry*, **168**, 1041–1049.
36. Maslach, C., Leiter, M.P. and Schaufeli, W. (2008) Measuring burnout. In: Cooper, C.L. and Cartwright, S. (eds.), *The Oxford Handbook of Organizational Well-being*. Oxford University Press, Oxford.
37. Ahola, K., Honkonen, T., Isometsa, E., Kalimo, R., Nykyri, E., Koskinen, S., Aromaa, A. and Lonnqvist, J. (2006) Burnout in the general population. Results from the Finnish Health 2000 study. *Soc. Psychiatry Psychiatr. Epidemiol.*, **41**, 11–17.
38. Kristiansson, K., Perola, M., Tikkanen, E., Kettunen, J., Surakka, I., Havulinna, A.S., Stancakova, A., Barnes, C., Widen, E., Kajantie, E. *et al.* (2012) Genome-wide screen for metabolic syndrome susceptibility loci reveals strong lipid gene contribution but no evidence for common genetic basis for clustering of metabolic syndrome traits. *Circ. Cardiovasc. Genet.*, **5**, 242–249.
39. Vartiainen, E., Laatikainen, T., Peltonen, M., Juolevi, A., Mannisto, S., Sundvall, J., Jousilahti, P., Salomaa, V., Valsta, L. and Puska, P. (2010) Thirty-five-year trends in cardiovascular risk factors in Finland. *Int. J. Epidemiol.*, **39**, 504–518.
40. Viitasalo, K., Lindstrom, J., Hemio, K., Puttonen, S., Koho, A., Harma, M. and Peltonen, M. (2012) Occupational health care identifies risk for type 2 diabetes and cardiovascular disease. *Prim. Care Diabetes*, **6**, 95–102.

41. Kivimaki, M., Lawlor, D.A., Davey Smith, G., Kouvonen, A., Virtanen, M., Elovainio, M. and Vahtera, J. (2007) Socioeconomic position, co-occurrence of behavior-related risk factors, and coronary heart disease: the Finnish public sector study. *Am. J. Public Health*, **97**, 874–879.
42. Kalimo, R., Hakanen, J. and Toppinen-Tanner, S. (2006) *Maslachin Yleinen Työuupumuksen Arviointimenetelmä MBI-GS [the Finnish Version of Maslach'sBurnout Inventory-General Survey]*. Finnish Institute of Occupational Health, Helsinki, Finland.
43. Maslach, C., Jackson, S.E. and Leiter, M.P. (1996) *Maslach Burnout Inventory Manual*. Consulting Psychologists Press, Palo Alto.
44. Taris, T.W., Schreurs, P.J.G. and Schaufeli, W.B. (1999) Construct validity of the Maslach burnout inventory-general survey: a two-sample examination of its factor correlates structure. *Work and Stress*, **13**, 223–237.
45. Leiter, M.P. and Schaufeli, W.B. (1996) Consistency of the burnout construct across occupations. *Anxiety Stress Coping*, **9**, 229–243.
46. Beck, A.T., Ward, C.H., Mendelson, M., Mock, J. and Erbaugh, J. (1961) An inventory for measuring depression. *Arch. Gen. Psychiatry*, **4**, 561–571.
47. Salokangas, R.K., Poutanen, O. and Stengard, E. (1995) Screening for depression in primary care. development and validation of the depression scale, a screening instrument for depression. *Acta Psychiatr. Scand.*, **92**, 10–16.
48. Karasek, R., Brisson, C., Kawakami, N., Houtman, I., Bongers, P. and Amick, B. (1998) The job content questionnaire (JCQ): an instrument for internationally comparative assessments of psychosocial job characteristics. *J. Occup. Health Psychol.*, **3**, 322–355.
49. Blin, N. and Stafford, D.W. (1976) A general method for isolation of high molecular weight DNA from eukaryotes. *Nucleic Acids Res.*, **3**, 2303–2308.